Part VII

Identity of System Controlling Expression of $\underline{a}_1^{m-1}$ and $\underline{a}_2^{m-1}$

During the summer of 1955 when tests of of the behavior of a_2^{m-1} were resumed, evidence was obtained indicating that an Spm-type element w was responsible for control of gene action at a_2^{m-1}. As pointed out in the last section, the expression of this Spm element was obscure and this contrasted with the clearity of this in the a_1^{m-1} cultures. However, several test crosses were made during the summer of 1955 the results of which suggested that the Spm element in the a_1^{m-1} cultures would control gene action at a_2^{m-1} ~~in as sharply~~ expressed a manner as it was known to do in the a_1^{m-1} cultures. Tests were then initiated to ~~determine if this could be~~ demonstrated ~~unambiguously~~. Some of these ~~tests~~ will be considered below.

During the summer of 1949, a plant that was homozygous for a_2^{m-1} had been crossed to one homozygous for the standard recessive, a_2. On the ear this cross produced, there weremany uniformly pale colored kernels and some variegated kernels that showed spots or specks of A_2 in a colorless background. In 1950, some of the kernels on this ear were sown under culture number 5303, and those in 5303C were derived from pale colored kernels in which there were one or several colorless areas within which specks of the A_2 phenotype were present. One plant

in culture 5303C, plant number 3, was variegated in that small streaks of the A_2 phenotype appeared in a non-pigmented background. This plant was a_2^{m-1}/a_2 in constitution. The first ear on the main stalk was self-pollinated. Among a total of 455 kernels this ear produced, 150 were uniformly pale colored, 195 were variegated, the pattern of ~~this~~ ranging from ~~one that had~~ spots of A_2 in a colorless background to tiny specks of A_2, either throughout the kernel or ~~in~~ restricted to colorless areas in an otherwise pale-colored kernel. In addition, there were 110 colorless kernels. An ear of a tiller of this plant was used in a cross with a plant homozygous for a_2. This ear was sectorial with regard to ~~the~~ expression ~~of~~ a_2^{m-1}. Nearly all the a_2^{m-1} kernels within a well defined sector, extending a distance of about 2/3rds of the ear and covering several rows, were uniformly pale colored. Only 3 kernels within this sector were variegated. They had tiny specks of the A_2 phenotype in a colorless background. On the rest of this ear, the a_2^{m-1} kernels segregated in a 1 : 1 ratio ~~of~~ uniformly pale colored to variegated. Among the variegated kernels, the pattern of A_2 mutant areas ranged from spots of A_2 in a colorless background in some kernels to specks of this in other kernels. Pollen of plant 5303C-2 was placed on the silks of an ear of a plant homozygous for a_2.

Among the a_2^{m-1} carrying kernels on the resulting ear, a ratio of approximating 1 uniformly pale colored kernel to 1 variegated kernels appeared and among the kernels in the variegated class the patterns of variegation were similar to those of the reciprocal cross, just described. Kernels from this ear were sown in the summer of 1955 under culture number 6908. Tests conducted with one plant in this culture will be described.

Plant 6903A-3 was derived from a pale colored kernels on the ear mentioned above.. ~~The plant proved to be variegated in that very small streaks of A_2 appeared in a non-pigmented background.~~ Pollen of this plant was placed on the silks of ears of three different plants, each homozygous for a_2. Two of these three plants ~~were~~ belonged to the regular a_2 tester stock. The third had quite a different history. It was derived from a colorless kernel on an ear produced by the cross of the the original plant carrying a_1^{m-1} to a plant that had a_2 but was homozygous for A_1. The original a_1^{m-1} carrying plant was a_1^{m-1}/a_1, A_2/a_2 in constitution and it carried a number of Spm elements. The colorless kernels on this ear should be a_2/a_2 and either A_1/a_1^{m-1} or A_1/a_1 in constitution, and ~~it~~ should have ~~several~~ Spm elements ~~in it~~. The plant ~~to which~~ used in the cross with plant 6903A-3 was ~~made proved to be~~ A_1/a_1, as later tests showed. The types of kernels on the ears

produced by crosses with the two a_2 tester plants and this a_2 plant were strikingly different. Among the 482 kernels on the ears produced by the former cross, all but 25 of the a_2^{m-1} kernels were uniformly pale colored. Of the 23 odd kernels, 11 were variegated for small specks of the A_2 phenotype in a colorless background and 12 were sectorial for such areas in an otherwise pale colored kernel. Among the 383 kernels produced by the latter cross, 183 were totally colorless and 200 were variegated, and the pattern of variegation was similar among all of the kernels expressing it. Spots of the A_2 phenotype appeared in a colorless background (Photo.). It was suspected that the Spm elements in the a_2/a_2 parent of this cross was responsible for the uniformly expressed variegated pattern in these kernels and that the Spm elements in it were the same as those that control gene action at a_1^{m-1}. Therefore, both the variegated kernels and colorless kernels on this ear were sown in the summer of 1956 under culture number 7113 and tests of the plants derived from them were commenced, in order to obtain evidence for the supposed identity of the system responsible for control of a_1^{m-1} and a_2^{m-1}.

Plants derived from variegated kernels on the ear just described were sown under culture numbers 7113A and 7113 B. Those derived from

the colorless kernels were sown under 7113C and 7113D. A number of different types of test were conducted with the plants of culture 7113 Included among them were crosses of the a_2^{m-1}/a_2 plants in A and B of culture 7113 to plants homozygous for a_2 derived from the tester stocks. ~~These tester plants were A_1/A_1 in constitution.~~ The types of kernels appearing on the ears produced by these test crosses are entered in table 1. Some of the plants in culture 7113 were also crossed by plants homozygous for a_1^{m-1} (state 5718) and for A_2 in which no Spm was present. This was done in order to determine the constitution of the plants in culture 7113 with respect to A_1 and also the presence in them of Spm elements capable of ~~altering~~ a_1^{m-1} in the ~~usual~~ manner. Some plants in culture 7113 proved to be A_1/A_1 and others proved to be A_1/a_1. In all plants of the latter phenotype, one or more active Spm elements were present as shown by the kernels on ears so produced, entered in table 2. Comparison of ratios of uniformly pale kernels (no Spm present) to variegated kernels (Spm present) for a_2^{m-1} and a_1^{m-1} ~~where this could be made~~ is summarized in table 3. ~~Conformity of~~ ratios ~~of types is evident~~.

In order to ~~expand~~ evidence ~~of~~ Spm control of both a_2^{m-1} and a_1^{m-1}, kernels were selected from ears of some of the crosses entered in table 2

and plants grown from them in the summer of 1957. These plants were tested in various ways, ~~now to be described~~. The phenotypes of thœ kernels that gave rise to the plants that were tested, as well as the number derived from each type, are shown in table 2 along with the 1957 culture number given to them. A description of these tests will commence with the plants in culture 7299, derived from the kernels on the ear produce by plant 7113C-5, table 2. From the ratio of pale colored to variegated kernels on this ear, it could be concluded that 7113C-5 carried one active Spm element. All plants derived from kernels on this ear should be a_1^{m-1}/a_1, A_2/a_2. Those derived from the uniformly pale colored kernels should have no active Spm element in them whereas those derived from the variegated class of kernels should contain an active Spm element. All plants derived from the pale class of kernels were themselves uniformly pigmented, indicating the absence of an active Spm element in them. All plants derived from the variegated kernels were variegated for small streaks of the A_1 phenotype in a non-pigmented background, indicating the presence in them of an active Spm element. To test for the ~~response of a_2^{m-1} to either~~ the presence or the absence of the Spm element that controls gene action at a_2^{m-1}, one or more ears of each plant were used in a cross with A_1/A_1 plants that were either a_2^{m-1}/a_2 or a_2^{m-1}/a_2^{m-1} and

in which no evidence of the presence of an Spm element was given either by the appearance of the plant or by the appearance of kernels on ears produced by crosses of them to a_2/a_2 tester plants. Among these a_2^{m-1} carrying plants, three different states were represented. One group of plants used as pollen parents in these crosses carried the class II state of a_2^{m-1} in one chromosome 5 and a_2 in the homologue. A second group of plants carried a class I state of a_2^{m-1} in one chromosome 5 and a_2 in the homologue. This state gives early occurring mutations to A_2 or a_2 in the presence of an active Spm element. The third group of plants carried this state of a_2^{m-1} in one chromosome 5 and another class I state in the homologue. This latter state gives many late occurring mutations to A_2 in the presence of an active Spm element.

Thirty ears were obtained from crosses of these a_2^{m-1} carrying plants to the 23 uniformly pigmented plants derived from the ear of plant 7113C-5 (table 2). On none of these ears did any variegated kernels appear. All the $a_2^{m-1}/a_2/a_2$ kernels exhibited either a full A_2 expression with the class II state of a_2^{m-1} or a uniformly pale pigmented phenotype with the class I states of a_2^{m-1}. In crosses of these same tester plants to the 18 variegated plants in culture 7299, thirty ears were obtained. On all ears, variegated kernels appeared. The $a_2^{m-1}/a_2/a_2$ kernels on

A fourth group of plants carried a class I state of a_{2n-1} gene chromosome 5 and a_2 in the homologue. This state produced a number of small spots of the A_2 phenotype and also some leaf tissue in the presence of an active Spm element (all plants -). This was also the state of a_{2n-1} that was present in two a_{2n-1} changes plants of culture 7113.

these ears were either uniformly pigmented or they were variegated for A_2 spots in a colorless background and the ratio of these two classes of kernels approached 1 : 1 on the majority of ears. ~~Gene action at both a_1^{m-1} and a_2^{m-1} was controlled by an Spm element in these plants. Thus, tests of the plants in culture 7299 supported the assumption that the~~ fully active Spm element in the a_1^{m-1} cultures was capable of controlling gene action at a_2^{m-1} and in the same manner that it does with a_1^{m-1}, ~~for~~ with the class I states of a_2^{m-1} this Spm induces mutation patterns that are quite consistent among the kernels carrying one particular state of a_2^{m-1}.

The same types of test as those just described were conducted with the progeny of plant 7113C-8 and 7113D-5, table 2. As shown in this table, the ratio of pale to variegated kernels on the ear produced by each of these two plants indicated that each of them carried more than one active Spm element. The results obtained from tests of the 43 progeny plants were similar to those just described, with the exception of two plants. The results obtained from tests of these two plants will be described shortly. Sixteen of the 17 plants derived from uniformly pale colored kernels were themselves uniformly pigmented. On ears produced by these 16 plants in crosses with the a_2^{m-1} tester plnats,

Thus, these tests showed that the response of a_1^{m-1} and a_2^{m-1} to the presence or absence of Spm in the same plant was alike. Those plants having an Spm capable of controlling gene action at a_1^{m-1} likewise had an Spm capable of controlling gene action at a_2^{m-1}. Conversely, those plants that had no Spm to which a_1^{m-1} would respond also had no Spm to which a_2^{m-1} would respond.

no variegated kernels appeared. All 26 plants derived from variegated kernels were themselves variegated for streaks of the A_1 phenotype in a non-pigmented background. The ears produced by 25 of these plants in crosses with the a_2^{m-1} tester plants indicated the presence in each of one or more active Spm elements capable of controlling gene expression at a_2^{m-1}. One fully active Spm element was present in 11 plants, two such elements were present in 8 plants and three or more were present in 6 plants, the number of Spm elements being ~~given by~~ the ratio of uniformly pale colored kernels to variegated kernels among these that were $a_2^{m-1}/a_2/a_2$ in constitution.

The two exceptional plants, mentioned above, are important for this study ~~for~~ the tests conducted with them and with their progeny serve to confirm the precise control of a_1^{m-1} and a_2^{m-1} behavior ~~by~~ and in ~~the same~~ manner ~~for each~~ by ~~one particular~~ Spm element. One of the plants, 7470A, derived from a uniformly pigmented kernel on the ear of plant 7113D-5 (table 2), had an Spm with altered action. It proved to be Spm-w that undergoes frequent but late occuring changes to Spm-s. The second exceptional plant, 7467-1, derived from a variegated kernel on the ear produced by plant 7113C-8 (table 2), carried an Spm element that was undergoing cyclical change ~~to the inactive~~ phase. The types of test conducted with these two plants and with their progeny

will be considered at the end of this section.

Plants 7113B-1 and B-9, table 2, were A_1/a_1, a_2^{m-1}/a_2 in constitution. From the ratio of pale to variegated kernels on the ears produced by the cross entered in this table, two or more active Spm elements were present in each. Tests conducted with the plants derived from the uniformly pale colored kernels and from the variegated kernels on these ears will now be considered. The 11 plants derived from the uniformly pale colored kernels were themselves uniformly pigmented. Fifteen test cross ears were obtained from these 11 plants, the type of cross being the same as that outlined above. On each of these ears, all $a_2^{m-1}/a_2^{s}/a_2^{s}$, $a_2^{m-1}/a_2^{m-1}/a_2$, or $a_2^{m-1}/a_2^{m-1}/a_2^{m-1}$ kernels were uniformly pigmented. None were variegated. Of the 8 plants derived from the variegated kernels (culture 7298, table 2) all were variegated. On the 10 testcross ears obtained from them, both uniformly pale colored kernels and variegated kernels appeared. From the ratio of these two phenotypes on the di ferent ears, it was concluded that one active Spm element was present in 5 plants, two active Spm elements were present in 3 plants, and two or more active Spm elements were present in 1 plant. Again, these tests showed that plants having no Spm as judged by a_1^{m-1} behavior had no Spm as judged by a_2^{m-1} behavior.

and those having Spm as judged by a_1^{m-1} behavior likewise had Spm as judged by a_2^{m-1} behavior.

Extensive tests were made with plants derived from the A_1, A_2 kernels entered in table 2. These kernels were selected from ears of plants in culture 7113 that had a_2^{m-1} in them. Each selected kernel had an area in it in which A_2, delivered by the male parent, had been lost. This allowed the presence of a_2^{m-1}, delivered by the female parent, to be revealed. In 17 of these kernels, this area had spots of A_2 in a colorless background, indicating the presence in the kernel of both a_2^{m-1} and of Spm. In one kernel, the a_2^{m-1} area was uniformly pale colored, suggesting that this kernel had received a_2^{m-1} from the female parent but no Spm element. Thus, all 18 plants derived from these selected kernels should be A_1/a_1^{m-1}, A_2/a_2^{m-1} in constitution and in 17 of them, Spm also should be present. Each of the 17 plants in which Spm was present were used in making crosses with plants homozygous for A_1 and for a_2 in which no Spm was present and also with plants that were homozygous for a_1 and A_2 and/or homozygous for a_1^{m-1} and A_2 in which no Spm was present. The phenotypes of kernels on the ears produced by test crosses of each of the 17 plants are entered in table 4, and the Spm constitution of each is summarized in table 5.

Only two of the 17 plants had one Spm element in them. All other plants had more than one ~~Spm element~~. In one of the two plants having one Spm element, plant 7296A-2, this Spm element ~~was~~ located close to a_2^{m-1} in chromosome 5. It will be noted that the ~~discrepancy in~~ ratio of uniformly pigmented kernels (no Spm) and variegated kernels (Spm present) is very ~~marked~~ different in the tests ~~of~~ a_1^{m-1} and a_2^{m-1}, ~~behavior~~. Plant 7296A-2 was a_2^{m-1} Pr/a_2 pr in constitution. The a_1^{m-1} tester plants used in making crosses with this plant was homozygous for pr. On these ears, the distribution of Pr and pr to the uniformly pale pigmented kernels and the variegated kernels indicated that the single Spm in plant 7296A-2 was located in the chromosome 5 carrying Pr and at a distance that was approximately 30 crossover units from Pr. The percent crossing over between Pr and A_2 is approximately 28 percent. Thus, the evidence from the two sets of test crosses placed Spm very close to a_2^{m-1} in plant 7296A-2.

Plant 7297A-2 had two Spm elements in it. It was Wx/wx and in the cross with the a_1^{m-1}/a_1^{m-1}, no Spm tester plant that also was wx/wx, linkage of one of the two Spm elements with Wx was made evident. Among the kernels ~~on the kernels~~ on the ear that were homozygous for a_1^{m-1} there were 111 pale colored kernels (no Spm) of which 27 were Wx and 84 were wx. Among the 288 variegated kernels, 167 were Wx and 121 were wx.

In tests of plant 7468A-2, evidence was given of the presence of 3 Spm elements in the cells that gave rise to each of the two ears tested of this plant. Pollen collected from a tassel of this plant was used on silks of ears of plants that were A_1/A_1, a_2/a_2 no Spm and also on ears of plants that were a_1^{m-1}/a_1^{m-1}, A_2/A_2 in which no Spm was present. The ratio of variegated to pale kernels on the ears these plants produced indicated that in the part of the plant that produced the tassel from which pollen was collected, only 2 Spm elements were present.

Plant 7298A, derived from an A_1, A_2 kernel in which an area was present that exhibited a pale phenotype was suspected to be A_1/a_1^{m-1}, A_2/a_2^{m-1} in constitution in which no active Spm element was present. It was used in crosses with plants that were A_1/A_1, a_2/a_2 in which no Spm was present and with plants that were a_1^{m-1}/a_1^{m-1}, A_2/A_2 in which no Spm was present. On the ears produced by these crosses, no variegated kernels appeared, A and C, table 6. In order to show that the a_2^{m-1} in this plant would respond to an active Spm element, the ear produced by the main stalk and an ear of a tiller were used in crosses with plants homozygous for A_1 and a_2 in which 1 Spm was present. (For tests of the presence of one Spm in each of these plants, 7308D-1 and 7308D-2, see page .) The types of kernels appearing on each of these ears is

given in B of table 6. It was clear from these tests that plant 7298A had no active Spm element in it to which either a_1^{m-1} or a_2^{m-1} could respond. It was also evident that the a_2^{m-1} in this plant would have responded to Spm had it been present. It was also known that the a_1^{m-1} delivered by the a_1^{m-1}, no Spm tester stock would have responded to Spm had it been present as these same tester plants had been unsed in many other crosses an these had clearly demonstrated its capacity to respond to Spm.

Altogether, 121 plants were tested in the summer of 1957 for the response of a_1^{m-1} and a_2^{m-1} to the presence or absence of Spm when the selection of plants to be tested for its presence or absence was based on the known response of only one of them, either a_1^{m-1} or a_2^{m-1}. In all tests, the response of a_1^{m-1} and a_2^{m-1} were quite alike to the presence or absence of Spm, and to the number of them, if present. There were no exceptions to this. The two plants among the 121, plant 6467-1 and 7470A-5, in which a single Spm with modified action was present, only served to strengthen the evidence of the common type of response of a_1^{m-1} and a_2^{m-1} to the same Spm element.

Plant 7467-1 was derived from a variegated kernel on the ear produced by plant 7113C-8, table 2. Its constitution was a_1^{m-1}/a_2, A_2/a_2^{m-1} (spotted). The plant derived from it was expected to be variegated for

A_1 streaks in a non-pigmented background. The main stalk of the plant, however, was uniformly pigmented as if no Spm were present in it but the tiller, however, was sectorial for areas in which small streaks of A_1 appeared in a non-pigmented background. The appearance of plant 7467-1 suggested that it had an inactive Spm element in it that was undergoing change to the active phase in some cells of the tiller. The silks of the first ear on the main stalk received pollen from a tester plant that was A_1/A_1, a_2^{m-1} (early) Bt/a_2^{m-1}(late) bt in which no Spm was present. Among the 393 kernels on this ear, 193 were fully pigmented, that is, A_2, and 200 were $a_2^{m-1}/a_2/a_2$. Among the latter, 133 were uniformly pale pigmented, as if no Spm were present in them, and 67 were variegated. However, in only 4 of these 67 variegated kernels was the type of variegati that expected to appear in the presence of a fully active Spm element. In the remaining 63 kernels, Spm was active only in a sector in the kernel (small specks of A_2 in a colorless area of an otherwise pale pigmented kernel) or was weakly active throughout the kernel (small specks of A_2 in a colorless background throughout the kernel).

The silks of the ear produced by the tiller received pollen from a plant that was A_1/a_1, a_2^{m-1}(spotted) Bt/a_2 bt in which no Spm was present. There were 156 kernels on this ear and 49 of them were fully pigmented

(A_1, A_2). In addition, there were 83 kernels that were either colorless or pale pigmented and 24 that were variegated. Among the latter, 7 were variegated for spots of A_1 in a colorless background, 11 had spots of A_2 in a colorless background, and 7 had a type of variegation that could be expected if the kernels had ~~neither A_1 or A_2 in them but only~~ a_1^{m-1} and a_2^{m-1}, ~~each~~ undergoing mutation to A_1 and A_2 respectively. The cross was made to obtain just these kernels in order to test the type of response of a_1^{m-1} and a_2^{m-1} to the ~~same~~ Spm element in the plants derived from ~~them~~ each.

From the ear of the main stalk, seven plants were grown from kernels that had small A_2 specks in a colorless background under culture number 7577A, and 3 plants were grown from the 4 variegated kernels in which a fully active Spm element appeared to be present under culture number 7577B. All 7 plants in culture 7577A were darkly pigmented except for a few areas in which pigment was absent, and in some of these areas, small streaks of the A_2 phenotype appeared. Obviously, these plants had an Spm element in them that was undergoing change in phase of activity, from inactive to active in some cells during the development of the plant. The appearance of the 3 plants in B of culture 7577 were similar. The plants in both A and B

of this culture were tested by crosses with plants homozygous for a_1^{m-1} (state 5719A-1) and A_2 in which no Spm was present and with plants homozygous for A_1 and a_2 in which no Spm was present. The appearance of the kernels on the ears these crosses produced is shown in table 7. It was obvious that the Spm element in each of these plants was behaving as it had done in the parent plant, 7467-1. It was in its inactive phase during much of the early development of the plant, turning to the active phase in some cells only late in development. The response of both a_1^{m-1} and a_2^{m-1} to the activity phases of Spm was exactly alike.

Four plants were grown under culture number 7578A from kernels on the ear of the tiller of plant 7467-1 that appeared to have Spm and and both a_1^{m-1} and a_2^{m-1} ($a_1^{m-1}/a_1^{m-1}/a_1^{m-1}$ or $a_1^{m-1}/a_1/a_1$ and $a_2^{m-1}/a_2/a_2$). In addition, six plants were grown under culture number 7578B from kernels that were uniformly pale pigmented, the type of pigment indicating that they were A_1, $a_2^{m-1}/a_2/a_2$ in constitution. The six plants in culture 7578B will be considered first. Five of them gave no evidence of the presence of Spm in them either from the appearance of the plant or from tests that were conducted with each. One plant, 7578B-4, however, had a few sectors in which small A_2 streaks appeared in a non-pigmented background. The remainder of this plant was uniformly pigmented.

It was obvious that this plant had an Spm element in it that was in its inactive phase in most parts of the plant, turning to the active phase in a few cells rather late in development. Thus, the pattern of Spm activity resembled that in the parent plant, 7467-1. The first ear of plant 7578B-4 which was A_1/a_1^{m-1}, a_2^{m-1}/a_2 in constitution, was used in a cross with a plant that was homozygous for A_1 and a_2 in which no Spm was present. On the resulting ear, there were 210 uniformly pale pigmented kernels, 215 totally colorless kernels, and one kernel that was variegated in part. Three-quarters of the aleurone layer of this kernel had spots of A_2 in a colorless background and one-quarter was uniformly pale pigmented. This was the only kernel on the ear that gave evidence of the presence of Spm in ~~the female parent~~. The Spm in this kernel had changed from the inactive to the active phase early in development of the endosperm. The second ear of the main stalk of plant 7578B-4 was used in a cross with a plant homozygous for a_1^{m-1} (state 5719A-1) and for A_2 in which no Spm was present. This ear produced 460 kernels, half of which were homozygous for a_1^{m-1}. In 79 of these latter kernels, evidence was given of the presence of Spm in them but in none of these kernels was Spm fully active. In most of them, Spm had changed from the inactive to the active phase rather late in development of the kernel.

Two other plants in culture 7578B were tested for the presence in them of an inactive Spm element by crosses of each with plants that were homozygous for A_1 and a_2 in which one or more active Spm elements were present. The usefulness of this test for revealing an inactive Spm element has been described elsewhere. An ear of plant 7578B-2 was used in a cross with a plant of the constitution given above in which one active Spm element was present (plant 7538-4, see page). On the resulting ear, there were 212 totally colorless kernels and 226 kernels carrying a_2^{m-1}. Among the latter, 152 were uniformly pale pigmented and 74 were colorless with spots of the A_2 phenotype and in nearly all of them large areas exhibiting the pale phenotype also were present. Had plant 7578B-2 carried an inactive Spm , half of the variegated kernels would have had no pale areas or only a few small pale areas. It could be concluded, then, that plant 7578B-2 had no Spm element in it. This same conclusion was drawn from a similar type of test of plant 7578B-3. An ear of this plant was used in a cross with plant 7538-6 which was homozygou for A_1 and a_2 and also for Spm (see page for discussion of the constitution of this plant). On the ear produced by this cross, 196 kernels were totally colorless and 186 carried a_2^{m-1}. Among the latter, 22 were uniformly pale pigmented and 164 had spots of A_2 in a colorless background. In the majority of the variegated kernels, large pale areas

also were present. Obviously, plant 7578B-3 had no Spm element in it. Thus, of the six tested plants in culture 7578B, only plant number 4 gave evidence of the presence of Spm in test crosses with either or both a_1^{m-1} and a_2^{m-1}.

Each of the four plants in culture 7578A had an Spm element in it. The kernels that gave rise to these plants were selected from the tiller ear of plant 7467-1 because the Spm element in each appeared to have undergone change in phase of activity considerably earlier than in the other Spm carrying kernels on this ear. If this change from inactive to active phase had occurred before gamete formation, an Spm with a new pattern of cyclical changes in phase of activity could be expected to appear in the plant derived from a kernel in which this had occurred. This proved to be ~~the case~~ for two of the four plants in culture 7578A. The tests conducted with each of these four plants is shown in table 8. In plants number 1 and 2, the Spm element in each underwent change in phase from inactive to active in a manner that resembled the pattern of this ~~given by it~~ in the parent plant, 7467-1. In plant number 3, Spm was active in most parts of the plant, undergoing change to inactive in a ~~few~~ cells, early in plant development. In plant number 4, Spm was active in all parts of the plant, undergoing change to inactive quite late in development, ~~and only in some cells~~.

The described tests of the progeny of plant 7467-1 clearly indicated that both a_1^{m-1} and a_2^{m-1} would respond ~~xxxxx~~ in like manner to ~~the activity phase of Spm and the~~ changes in ~~this~~ that occurs in a plant or a kernel. Thus, these tests serve as an additional proof of the identity of the system controlling gene action at a_1^{m-1} and a_2^{m-1}. Similar evidence was obtained from tests of the progeny of plant 7470A-5, to be described below.

Plant 7470A-5 was derived from a uniformly pale pigmented kernel on the ear produced by the cross of plant 7113-5 (table 2). It carried an Spm element but the action of this elementswas weak (Spm-w) in most parts of the plant. Change from Spm-w to Spm-s occurred in some cells but only late in development. of the plant and of the endosperm tissue. This plant was a_1^{m-1} (state 5718)/ a_1, A_2/a_2 in constitution. The first ear of the main stalk was used in a cross with a plant that was A_1/A_1, a_2^{m-1}/a_2 and in which no Spm was present. The state of a_2^{m-1} was one that gives early occurring mutations to A_2 and to colorless in the presence of a fully active Spm element. On the ear this cross produced, there were 127 kernels that were uniformly deeply pigmented (A_2) and 69 that were totally colorless ($a_2/a_2/a_2$). There were 64 kernels that were $a_2^{m-1}/a_2/a_2$ in constitution. Thirty-three of them were uniformly pale pigmented and

31 were variegated. ~~The~~ ~~In~~ In only one of the latter kernels was the pattern of variegation that which appears when a fully active Spm is present (see photo.)/ In all other kernels, only small spots of the A_2 phenotype appeared in a colorless background (photo.). Nine plants were grown from these kernels under culture number 7580A. The single kernel that had large A_2 and colorless areas (~~Spm-s~~ variegation pattern with Spm-s) was sown under culture number 7580B. This plant, however, died during the very unfavorable growing condutions of the spring of 1958.

The types of test cross conducted with the plants in culture 7580A are shown in table 9. Each had an Spm-w element in it. Both a_1^{m-1} and a_2^{m-1} responded ~~in~~ like manner to this Spm-w element. The state of a_1^{m-1} used in the test cross gives only one or a few spots or specks of A_1 or none at all in the presence of Spm-w. In the presence of Spm-s, it gives many small spots of A_1 (compare photo. with photo.). The state of a_2^{m-1} responds to Spm-w by producing a number of small spots of A_2 in a colorless background. With Spm-s, it gives many large areas of A_2 as well as a number of smaller areas (compare photo. with photo.). On all testcross ears, some kernels were present ~~in which~~ with sectors ~~appeared~~ within which a pattern of variegation appeared resembling that produced by Spm-s

(photos). The time during development of change of Spm-w to Spm-s was so late that very few kernels were produced that exhibited an Spm-s type of response of a_1^{m-1} or a_2^{m-1} throughout the kernel. Nearly all such changes occurred during endosperm development, and the majority of these took place rather late in its development. Evidence of this was given by the response of both a_1^{m-1} and a_2^{m-1} to these changes in degree of activity of Spm.

The ear of the tiller of plant 7470A-5 was used in a cross with a plant that was A_1/a_1, a_2^{m-1}(spotted)/a_2 in which no Spm was present. On the ear this cross produced, there were 58 fully pigmented kernels (A_1 and A_2), 133 colorless kernels, 32 lightly pigmented kernels and 62 kernels tha had spots of deep pigmentation in a colorless background. Among the variegated kernels, 10 appeared to be a_1^{m-1}, A_2 in phenotype, 11 appeared to be A_1, a_2^{m-1} in phenotype and 41 were nearly totally colorless, only a small spot or speck of deep pigment being seen in these kernels. Six plants were grown from the variegated kernels that appeared to be a_1^{m-1}, A_2 under culture number 7581A and five plants were grown from the kernels that were nearly colorless, under culture number 7581B. All plants in A of this culture were variegated in that some small streaks of deep anthocyanin pigment appears in a non-pigmented background. The plants in

B of this culture were nearly completely non-pigmented. Only an occasiona very small streak of anthocyanin pigment appeared in them. The test crosses conducted with the plants in culture 7581 and the results obtained from them are entered in table 10. In three of the five tested plants in A of this culture, one Spm-w element was present. It was behaving much as it had done in the Spm carrying parent plant. Changes from Spm-w to Spm-s were occurring but only late in development. Plants 7581A$_{-1}$ and A-4 had 2 Spm elements in then. One of these had the Spm-w state and the other was Spm-s. This latter appeared to be undergoing change in phase of activity from inactive to inactive in some cells, late in development.

The four tested plants in B of culture 5781 all had an Spm-w element i them and it was behaving much as it had dome in the parent plant. These plants probably were a_1^{m-1}, a_2^{m-1} although tests for this were conducted only with plant B-4. The Spm-w in these plants was changing to Spm-s in some cells of the kernels having it but usually, only late in developmen of the kernel. In plant 5781B-4, tests were conducted of the response of a_1^{m-1} and a_2^{m-1} to the Spm in it. The states of each were much alike and the pattern of spots each produced in the kernel in the presence of the Spm in this plant was much alike. Each produced only a few deeply pigmented spots in the presence of the Spm-w element in this plant. Also, each responded in like manner to change of Spm-w to Spm-s.

With both a_1^{m-1} and a_2^{m-1}, Spm-w reduces the frequency of occurrence of mutation in comparison to Spm-s and also retards the time of their occurrence. With the two a_2^{m-1} states examined (early and spotted) the effect produced by Spm-w was the same as that which given by similar states of a_1^{m-1}, reviewed in section IV.

The common response of a_1^{m-1} and a_2^{m-1} to the same Spm elements was revealed by other tests conducted during the summer of 1958. In all cases, both a_1^{m-1} and a_2^{m-1} responded in like manner to the Spm element or elements in a tested plant. Since the tests reported in this section reveal this common response, the additional tests need not be reviewed here